A Study on the Structure and Morphologic Development of Calcaneal Tendon and Triceps Surae Muscle in Human Fetuses during the Fetal Period and the Evaluation of Clinical Importance of Calcaneal Tendon

Estudio sobre la Estructura y Desarrollo Morfológico del Tendón Calcáneo y el Músculo Tríceps Sural en Fetos Humanos Durante el Período Fetal y Evaluación de la Importancia Clínica del Tendón Calcáneo

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SUMMARY: It was aimed that the morphometric development of calcaneal tendon and the structures building it up in human fetuses during the fetal period be anatomically studied and that its clinical importance be evaluated. The study comprised a total of 102 fetus legs (51 human fetuses: 26 male and 25 female) whose ages varied between 15-40 gestational week, without external pathology or anomaly. The fetuses were divided in groups according to gestational weeks, trimesters and months. In the wake of the general external measurements of fetuses, leg dissection was performed. Afterwards, the morphometric parameters of gastrocnemius muscle, soleus muscle and calcaneal tendon were measured. The averages and the standard deviations of the measured parameters were determined according to gestational weeks, trimesters and months. There was a significant correlation between the measured parameters and the gestational age (p<0.001). There was no difference between sexes in terms of parameters (p>0.005). All the obtained results were discussed by making a comparison between them and the previous studies made. We are of the opinion that the data obtained in our study will be of use to the involved clinicians in the evaluation of the development of calcaneal tendon and the structures constituting it during the fetal period and in clinical studies and applications as well.

KEY WORDS: Fetal development; Morphometry; Calcaneal tendon; Anatomy.

INTRODUCTION

Identified sonographically for the first time by Fornege in 1986, calcaneal tendon is formed of gastrocnemius muscle and soleus muscle tendons which connect in the mid-region of the leg. Separately, plantaris muscle may also participate in the formation of calcaneal tendon (Doral *et al.*, 2010; Grechenig *et al.*, 2004; O'Brien, 2005; Pichler *et al.*, 2007; Standring *et al.*, 2008). The length of the tendon in adults is approximately 15 cm. (Harris & Peduto, 2006; Standring *et al.*, 2008). Originating from the mid-regions of the leg, the tendon ends up at the tuber calcanei in the calcaneus bone or heel bone in the Tarsus (Harris & Peduto, 2006; Standring *et al.*). Separately, the course of direction of the fibres of calcaneal tendon in adults are stated to be spiral (Apaydin *et al.*, 2011; Grechenig *et al.*; Standring *et al.*). While calcaneal tendon receives a blood supply from the peroneal artery and posterior tibial artery, it also receives stimulation from tibial and sural nerves (Doral *et al.*). These muscles and calcaneal tendon are located superficially at the posterior region of the calf of the leg. Gastrocnemius and soleus muscles together form the triceps surae muscle. Gastrocnemius muscle has a fusiform and two heads called caput mediale and caput laterale. The broad medial head originates from the popliteal surface right above the medial condyle of femur. The lateral head, on the other hand, originates from the posterolateral surface of the lateral condyle of femur and from the lateral lip right below

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linea aspera. Soleus muscle is located below gastrocnemius muscle. It is broad, flat, bipennate or multipennate muscle was named because of its resemblance to sole, a flat fish (Doral *et al.*; Moore, 1992; O'Brien; Pichler *et al.*; Standring *et al.*).

Calcaneal tendon is the strongest and the largest tendon of the human body (Doral et al.; Grechenig et al.; O'Brien). It is principally responsible for the plantar flexion of the ankle (O'Brien; Pichler et al.; Standring et al.). Separately, having a significant characteristic in strength conduction, calcaneal tendon is of a rigid and flexible character (Doral et al.). During running, jumping and skipping, calcaneal tendon is exposed to strain ten times as much as the body weight and can lengthen by 4% more before sustaining any injury and therefore, tendon ruptures commonly occur (Doral et al.; O'Brien). It is expressed that tendon ruptures occur more in males due to the fact that calcaneal tendon is of a rigid character in them (Doral et al., 2010). Besides, it is stated in studies that achilles tendinopathy occurs commonly in athletes (Zafar et al., 2009). It is pointed out that there is a 52% of risk for tendinopathy, particularly in the long-distance male runners throughout their lives (Zafar et al.). It is stated that in the treatment of achilles tendinopathy, there is 29 % requirement for surgery (Zafar et al.). Separately, it is explained that the blood build-up in the mid-region of the tendon is insufficient, due to which several problems occur in this region (Doral et al.).

There have been many radiological, clinical and anatomic studies in adults with respect to the morphometry of achilles tendinopathy (Greenhagen et al., 2012; Kim et al., 2010; Talsnes & Sudmann, 1992). Doral et al., in a study they conducted, studied and analyzed the achilles tendon functionally and anatomically. In another study, the researchers researched into the underlying reasons for achilles tendinopathy and its clinical importance (Zafar et al.). Harris & Peduto in their studies, intended to create an archive by radiologically imaging the normal and diseased achilles tendons. Additionally, in a number of conducted studies, the researchers aimed at finding the thickness of achilles and patella tendons through the ultrasonographic method (Fredberg et al., 2008; Kallinen & Suominen, 1994; Koivunen-Niemelä & Parkkola, 1995). Chao et al. (1997) researched into the termination positions of the achilles tendons anatomically in fresh cadavers. Separately, Pichler et al. examined the termination points of soleus muscle in calcaneal tendon and analyzed its clinical importance in adult cadavers. In another study, Bénard et al. (2011) analyzed the effect of growth in children on the geometry of gastrocnemius muscle through the ultrasonographic method.

Different from other ones, in our study, we aimed at researching anatomically into the morphometric development of calcaneal tendon, and gastrocnemius muscle caput mediale/laterale and soleus muscle joining the constitution of this tendon throughout the fetal period by using the anatomic dissection method on human fetuses, and we also intended to discuss its importance in terms of clinical aspects.

MATERIAL AND METHOD

The study comprised of 102 fetus legs (51 human fetuses: 26 male and 25 female) at a gestational age of 15-40 weeks; the fetuses were obtained from the prenatal period or after abortion. All were spontaneous abortions or stillbirths and neonatal deaths (died owing to premature or prenatal asphyxia) obtained from Isparta Maternity and Paediatric Hospital during 1996–2012. In order to use the fetuses as experimental materials, the signed consents were obtained from the families and the experimental procedures were ethically approved by the official laws and regulations of Turkish Ministry of Health. The fetuses with external pathology or anomalies and those cases with anomalies (omphalocel, gastroschisis, diaphragm hernia, Meckel diverticulum, colon malposition, renal agenesis, ectopic kidneys, agenesis of external genitalia, etc.) after dissection were also not studied.

Gestational ages of the fetuses were determined using crown-rump length (CRL), bi-parietal diameter (BPD), head circumference (HC), femur length (FL) and foot length (Moore & Persaud, 2009). Fetuses were assigned to one of three groups according to their gestational ages: Group I (2nd trimester, 15-25 weeks), Group 2 (3rd trimester, 26–37 weeks) and Group 3 (term, 38–40 weeks). Fetuses were also divided into 7 groups according to their gestational age in months; fetuses aged 13–16, 17–20, 21–24, 25–28, 29–32, 33–36, and 37–40 weeks were assigned to 4, 5, 6, 7, 8, 9, and 10-months groups, respectively.

First of all, in all the fetal materials, leg dissection was performed through the anatomic dissection method. By removing the cutaneous and subcutaneous fat layer in the posterior region of the leg, the sural nerve, gastrocnemius muscle caput mediale/laterale, soleus muscle and calcaneal tendon in the posterior leg were made visible. Later, the morphometric measurements of the muscles and calcaneal tendon were taken. The measurements pertaining to the macroscopic parameters used in the study were taken with the help of digital Insize calipers.

The Parameters Taken During The Study

Length of the leg (a): The vertical distance stretching between the intermalleolar line and the transverse axis passing along the medial region of the knee joint.

Length of gastrocnemius muscle caput mediale (b): The vertical distance between the transverse axis passing along the initial starting point of the muscle and the transverse axis passing along the initial point of the calcaneal tendon.

Width of gastrocnemius muscle caput mediale (c): The largest transverse distance between the vertical axes passing along the lateral and medial sides of the muscle in the way that it will exist in the medial region of the muscle.

Thickness of gastrocnemius muscle caput mediale (d): The thickest sagittal distance between the transverse axes passing along the frontal and posterior surfaces of the muscle in the way that it will exist in the medial region of the muscle.

Length of gastrocnemius muscle caput laterale (e): The vertical distance between the transverse axis passing along the initial point of the muscle and the transverse axis passing along the initial point of the calcaneal tendon.

Width of gastrocnemius muscle caput laterale (f): The largest transverse distance between the vertical axes passing along the lateral and medial sides of the muscle in the way that it will exist in the medial region of the muscle.

Thickness of gastrocnemius muscle caput laterale (g): The thickest sagittal distance between the transverse axes passing along the frontal and posterior surfaces of the muscle in the way that it will exist in the medial region of the muscle.

Length of soleus muscle (h): The vertical distance between the transverse axis passing along the initial point of the muscle and the the transverse axis passing along the initial point of the calcaneal tendon.

Width of soleus muscle (1): The largest transverse distance between the vertical axes passing along the lateral and medial sides of the muscle in the way that it will exist in the medial region of the muscle.

Thickness of soleus muscle (i): The thickest sagittal distance between the transverse axes passing along the frontal and posterior surfaces of the muscle in the way that it will exist in the medial region of the muscle.

Length of calcaneal tendon (j): The vertical distance between the transverse axes passing along the initial and termination points of the tendon.

Initial width of calcaneal tendon (k): The largest transverse distance between the vertical axes passing along the medial and lateral sides in the initial region of the tendon.

Termination width of calcaneal tendon (l): The largest transverse distance between the vertical axes passing along the medial and lateral sides in the termination region of the tendon.

Initial thickness of calcaneal tendon (m): The thickest sagittal distance between the transverse axes passing along the frontal and posterior surfaces in the initial region of the tendon.

Termination thickness of calcaneal tendon (n): The thickest sagittal distance between the transverse axes passing along the frontal and posterior surfaces in the termination region of the tendon.

Later on, histological tissue samples were taken from the soleus and gastrocnemius muscles and calcaneal tendon for the microscopic analysis. Each tissue sample was embedded in paraffin blocks in a vertical and coronal (for the tendon) way after the routine histological tissue followup methods were applied. Then, histological sectioning was done from the proximal region to the distal region of the tendon in a vertical and coronal position until the tissue samples with 5 micrometer thickness were used up. Through the systematic random sampling method, 7–10 pieces of sample sections were taken from each case. The obtained sections were dyed by means of Hematoxylin and Eosin (H&E) and were evaluated under the light microscope (Olympus CX41RF).

Statistical Analysis. By utilizing the SPSS statistical program, the averages and standard deviations of the parameters according to genders, gestational age and groups were ascertained. The significance level in the statistical analysis was taken as p<0.05. The parametric values given in accordance with the groups were shown with the average \pm standard deviation. In the comparison of the groups, nonparametric tests were used due to the scarcity of the number of cases in some groups. Firstly, the Kruskall-Wallis variance analysis was performed. As the result of this analysis, the groups regarded as significant were compared in groups of two by means of Mann-Whitney U test. The levels of significance were assessed through the Bonferroni Correction. The relationships between the parameters taken and the gestational age (week) were determined through the use of Pearson correlation test. In the inter-gender comparison of the parametric data, the Student- T test (in total for all the cases) and Mann-Whitney U test (within each group while comparing separately) were utilized. The values, p obtained were given in the findings section and under the tables involved.

RESULTS

First of all, the general external measurements of fetuses were taken. Then the posterior leg dissection was performed. By removing the cutaneous and subcutaneous fat layer in the posterior region of the leg, the sural nerve, gastrocnemius muscle caput mediale and caput laterale, soleus muscle and calcaneal tendon in the posterior leg were made visible (Fig. 1A-B). The neighbourhood relations of calcaneal tendon with the neighbouring structures at the posterior leg was examined, and the neighbouring relations were observed to be normal (Fig. 1A-B). Afterwards, the morphometric measurements of the leg length of fetuses, gastrocnemius muscle caput mediale and caput laterale, soleus muscle and calcaneal tendon were taken. The averages and standard deviations of these measurements were determined according to weeks, trimesters and months (Tables I, II and III). The measured parameters had a significant correlation between them and the gestational age (p < 0.001). There was no difference between the genders in terms of parameters (p>0.005). In the comparison of these parameters made among the trimester groups; there was no difference among the other groups (p<0.05, Table II), except for the 3rd and 4th trimesters in the parameter of calcaneal tendon length (j) and the 3rd and 4th trimesters in the parameter of calcaneal tendon initial thickness (m). On the other hand, in the comparison of these parameters made among the months; there was a difference among the other groups (p<0.05, Table III) except: in the parameter of leg length (a) between the 4.-5., 4.-6., 5,-6., 6.-7., 7.-8., 8.-9., 9.-10th months ; in the length parameter of gastrocnemius muscle caput mediale (b) between 4.-5., 8.-

Table I. The averages of the parameters of calcaneal tendon according to weeks (mm).

u	0.38	0.46	0.51	0.59	0.67	0.90	0.94	1.00	1.08	1.35	1.86	2.11	2.53	2.97	3.23	3.66	4.12	4.62	5.02	5.58	6.03	6.49	6.99	7.19	7.38	7.93
m	0.47	0.65	0.72	0.80	0.85	1.08	1.29	1.55	1.73	2.29	2.65	2.97	3.30	3.77	4.10	4.44	4.84	5.13	5.75	6.07	6.51	6.96	7.27	7.59	7.89	8.09
Ι	0.86	1.18	1.33	1.53	1.86	2.09	2.48	2.69	2.95	3.27	3.77	4.28	4.82	5.51	6.65	7.48	8.20	8.89	9.53	10.29	11.38	11.91	12.36	12.92	13.18	13.86
k	1.21	1.74	1.92	2.15	2.37	2.67	2.99	3.30	3.75	4.12	4.45	5.17	6.43	7.00	7.87	8.18	9.15	66.6	10.37	11.44	12.28	12.88	13.18	13.69	14.01	14.40
· - ,	6.41	6.96	7.35	8.04	8.62	9.27	10.08	11.27	12.81	13.63	14.21	15.51	16.28	17.95	19.10	20.94	21.71	22.81	24.18	25.89	27.28	28.57	29.21	30.10	31.16	31.97
•=	1.02	1.11	1.18	1.44	1.48	1.96	2.30	2.57	2.89	2.96	3.17	3.31	3.57	4.07	4.47	5.19	5.67	6.21	7.02	7.58	7.98	8.14	8.50	8.83	9.14	9.44
I	4.98	5.39	6.09	6.51	7.08	7.90	8.36	9.18	10.03	10.75	11.66	12.50	13.26	13.79	14.92	15.66	16.56	17.98	18.59	19.23	20.66	22.24	23.82	24.26	25.30	25.67
ų	16.57	17.42	18.51	19.65	20.57	21.78	22.42	23.65	24.51	26.51	28.48	31.26	33.63	36.65	38.30	39.79	41.32	43.60	45.16	47.52	49.53	51.66	52.52	53.53	54.22	55.76
5.0	0.65	0.79	1.01	1.11	1.20	1.33	1.47	1.75	1.96	2.05	2.21	2.59	2.89	3.03	3.24	3.78	4.08	4.21	4.75	5.13	5.86	6.11	6.40	6.98	7.19	7.33
f	3.20	3.54	3.88	4.17	4.78	5.15	5.75	6.30	6.61	7.21	7.86	8.16	8.94	9.48	9.75	10.34	10.83	11.55	12.06	12.86	13.37	14.03	14.55	15.09	15.73	16.17
e	13.50	14.51	15.27	16.38	17.90	18.39	20.90	21.53	22.95	24.38	25.66	27.70	29.51	32.01	34.15	36.19	38.97	40.65	42.02	44.14	45.85	47.30	48.07	49.83	50.59	51.83
q	0.78	0.88	1.11	1.28	1.30	1.39	1.95	2.11	2.14	2.17	2.25	2.94	3.27	3.52	3.87	4.09	4.80	5.06	5.78	6.20	6.70	6.93	7.07	7.15	7.44	7.57
c	4.07	4.18	4.30	4.57	5.24	5.99	6.78	6.87	7.25	8.15	8.56	9.44	9.80	9.98	10.44	11.24	11.43	12.58	13.43	13.62	14.21	15.04	15.21	16.20	16.53	16.88
q	14.77	15.79	16.89	17.63	19.55	20.17	21.75	22.87	23.63	25.75	27.72	30.06	32.30	35.94	37.24	38.92	40.57	42.48	44.59	46.35	47.40	49.41	50.74	51.67	52.97	53.54
а	28.01	30.31	33.61	35.97	38.98	40.71	43.80	46.53	48.41	50.93	52.09	54.23	56.78	58.54	60.69	63.67	65.99	67.93	69.06	71.89	72.30	74.61	76.82	78.28	79.40	80.89
u	2	0	4	4	4	4	4	4	9	9	4	4	4	9	7	9	4	4	4	4	4	7	4	4	0	4
Age	15. we eks	16. we eks	17. we eks	18. we eks	19. we eks	20. we eks	21. we eks	22. we eks	23. we eks	24. we eks	25. we eks	26. we eks	27. we eks	28. we eks	29. we eks	30. we eks	31. we eks	32. we eks	33. we eks	34. we eks	35. we eks	36. we eks	37. we eks	38. we eks	39. we eks	40. we eks

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Table II. The	average	es and stand	ard deviatio	ns of calca	neal tendo:	n parametei	rs accordin	ig to the tr	imesters (g	troups) (m	m).					
Groups (trimesters)	=	æ	q	3	p	ల	f	60	-	-		· ··	¥	-	E	-
Group 1 (15-25 weeks)	44	40.2±9.2	22.4±5.9	6.2±1.9	1.5±0.5	22.4±6.6	5.8±1.9	1.4±0.4	30.1±6.6	9.1±2.8	1.8±0.7	11.3±3.7	2.8±1.1	1.9 ± 0.6	0.9±0.3	0.7±0.2
Group 2 (26-37 weeks)	48	61.8±10.8	35.5±4.8	10.7 ± 1.7	2.8±0.7	36.6±5.2	10.5±1.8	2.4±0.7	43.3±5.2	16.4±3.	5 3.4±1.0	23.2±5.2	6.6±2.1	3.7±0.8	1.7±0.5	1.3 ± 0.3
Group 3 (38-40 weeks)	10	75.3±23.9	47.4±2.7	14.7±1.5	3.7±0.7	46.9±5.1	14.1±1.5	3.4±0.9	56.4±5.1	22.2±1.) 4.7±1.5	28.3±4.4	8.3±1.4	4.7±0.4	1.9 ± 0.4	1.7±0.2
T otal (15-40 weeks)	1 02	53.8±17.2	31.0±9.7	9.1±3.3	2.3±0.9	31.3±10.2	8.8±3.3	2.1±0.9	38.9±10.3	13.8±5.	4 2.8±1.3	18.6±7.9	5.2±2.6	3.0±1.2	1.3±0.6	$1.1 {\pm} 0.4$
P<0.05= Diffe 3rd and 4th trii Table III. The	ence bet nesters.	ween trimest	ers (calcaneal lard deviation	l tendon in the	ie length pai	rameter excej	pt for (j): be	stween 3rd ^ε ng to mon	and 4th trime ths (proures	esters; canea	ıl tendon in	the initial th	ickness pa	rameter exc	cept for (m)	between
Groups (months)	=	a	q	J	p	ووال	f	ac	4	-			k	-	E	=
4. month	4	29.4±0.8	16.3±1.1 4	.4±0.4 0	.9±0.1	14.0±1.6	3.2±0.4	0.9 ± 0.1	21.6±1.7	5.5±0.8	1.2 ± 0.1	6.8±0.8	1.5±0.4	1.0±0.2	0.6±0.1	0.5±0.1
5. month	14	33.4±5.8	18.0±3.3 4	.6±1.1 1	.2±0.3	17.6±3.0	4.3 ± 1.3	1.1 ± 0.2	25.0±3.8	6.8±1.5	1.3 ± 0.4	9.0±0.2	2.1±0.4	1.4 ± 0.3	$0.7{\pm}0.1$	0.6±1.1
6. month	20	43.8±7.1	25.2±5.1 7	.1±1.7 1	.8±0.4	25.2±5.5	6.6±1.5	1.6 ± 0.3	33.9 ± 4.9	10.4 ± 2.0	2.1 ± 0.7	12.2±2.9	3.1 ± 1.0	2.1±0.5	0±0.37	$0.7{\pm}0.1$
7. month	20	53.6±3.8	30.2±3.5 8	.9±1.1 2	.1±0.2	31.4±4.2	8.5±1.0	1.9 ± 0.4	38.0±4.6	13.6±1.6	2.6 ± 0.8	17.9±2.6	4.7±1.2	2.9±0.5	1.3 ± 0.3	1.0 ± 0.1
8. month	16	59.9±14.6	35.5±3.4 10	0.4±1.7 2	.7±0.5	36.7±4.2	10.4 ± 1.2	2.2±0.5	44.1 ±4.0	$16.1 {\pm} 2.1$	2.9±0.7	23.7±6.0	6.9±2.4	3.5±0.6	1.7 ± 0.4	1.3 ± 0.2
9. month	12	67.6±4.6	37.8±5.0 1	1.4±1.0 3	.1±0.4	38.3±4.6	11.7 ± 1.1	2.8 ± 0.4	45.9±4.9	17.1±4.8	3.8±0.7	26.8 ± 1.4	8.0±0.8	4.3±0.7	1.9 ± 0.2	1.5±0.3
10. month	16	74.2±18.7	45.1±4.4 1.	4.1±1.7 3	· 3±0.8	44.6±5.6	13.4 ± 1.9	3.4 ± 0.8	53.9±5.7	21.9±2.3	4.7±1.3	27.4±4.3	8.0±1.5	4.7±0.4	2.0±0.5	1.6±0.2
P<0.05= Diffe mediale (b) bet mediale (d) bet caput laterale ((h) between 4 8. 78., 89., 46., 56., 8', 5., 46., 67.,	ence bet ween 4 ween 4 () betwee 5., 67., 910th n)., 810. 89., 8	ween months 5., 89th mor -5., 46., 67 an 4.5., 89th month nonths; in the , 910th mon 10., 910th m	(in the pararr 10, 8, -9th mont ., 8, -9th mont months; in the s; in the widt length pararr ths; in the ter ths; in the ter nonths; and a	teter of leg le didth paramet hs; in the ler e thickness p h parameter neter of calcar mination wic lso except in	ngth, exclut er of gastroc ugth parame arameter of of soleus mu neal tendon the termina	ding: (a) betw memius musk ter of muscul gastrocnemii gastrocnemi uscle (1) between (j) between er of calcane: tion thickney	/een the 4 $\frac{1}{2}$ caput me cle caput me ulus gastrocon us muscle cc /een 45., 7 45., 46., al tendon (1) ss parametel	 5., 46., 5,-6 ediale (c) be ennius capu aput laterale aput laterale 56, 89., 8 between 4. r of calcane 	5., 67., 78 tween 45., t laterale (e) (g) betweer months; in tl 310, 910th 5., 910th al tendon (n	., 89., 91 78., 89th between 4 1.45., 46., a thickness th months; in months; in months; in	Dth months; in months; in months; in $5., 89$ th m $47., 67.$, parameter of parameter of in the initial the ininitial the ininitial the initial the initial	in the lengt the thickness on this; in the on this; in the of the mont of soleus mu width paran ickness para: -6., 89., 8	h paramete s paramete v width par hs;in the le hs:le (i) be neter of cal umeter of c 10., 91(r of gastroc r of gastroc ameter of g ngth param tween 45. caneal tend alcaneal ten bth months)	nemius mu nemius mu astrocnemi eter of sole (46., 56. on (k) betw ndon (m) be	scle caput scle caput is muscle 67., 6 een 45., tween 4
tendon (n) between 45., 4 6., 56., 89., 810., 910th months.	termination thickness parameter of calcaneal	of calcaneal tendon (m) between 45., 46., 67., 8 9., 810., 910th months; and also except in the	calcaneal tendon (l) between 45., 910th months; in the initial thickness parameter	6., 56., 89., 810., 910th months; in the termination width parameter of	months; in the initial width parameter of calcaneal tendon (k) between $4 - 5 - 4$	10th months; in the length parameter of calcaneal tendon (j) between 45., 4	parameter of soleus muscle (i) between 45., 46., 56., 67., 68., 78., 89., 9	parameter of soleus muscle (1) between 45., 78., 8 9th months; in the thickness	parameter of soleus muscle (h) between 45., 67., 8 9th months; in the width	laterale (g) between 45., 4 6., 47., 67., 78th months; in the length	between 4.5., 89th months; in the thickness parameter of gastrochemius muscle caput	months; in the width parameter of gastrocnemius	parameter of gastrocnemius muscle caput laterale (e)	muscle caput mediale (d) between 45., 46., 67., 8 9th months: in the length	between 45., 78., 89th months; in the thickness parameter of gastrocnemius	9th months; in the width parameter of gastrocnemius muscle caput mediale (c)

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Fig. 1. A) From the postero-medial aspect of the leg. 1= Calcaneal tendon, 2= Tibialis posterior artery and vein, 3= Flexor digitorum longus muscle, 4: Tibialis posterior muscle, 5= Medial malleol, 6= Gastrocnemius muscle, 7= Calcaneus. B) From the postero-lateral aspect of the leg. 1= Calcaneal tendon, 2= Sural nerve, 3= Fibularis longus and breves muscles, 4= Lateral malleol, 5= Soleus muscle, 6= Gastrocnemius muscle, 7= Calcaneus.

DISCUSSION

It is stated that the pathologies regarding the calcaneal tendon seen in adults is important (Apaydin et al.; Greenhagen et al.). In fact, the pathologies, such as the shortness of tendons, related to calcaneal tendon are said to have started to occur during the fetal period (Greenhagen et al.). Separately, it is pointed out in the studies that there are changes taking place in the formation of calcaneal tendon in pathologies, such as Haglund's deformity, retrocalcaneal bursitis, rupture and enthesopathies, and that such cases cause clinical problems concerned with walking (Apaydin et al.; Greenhagen et al.). For this reason, it is important to know the anatomy of calcaneal tendon right from the start of the fetal period. Considering the previous studies conducted on calcaneal tendon, no data is found in regard to the fetal development of calcaneal tendon. In our study, different from previous ones, it was aimed that more detailed morphometric data regarding the fetal calcaneal tendon be obtained by using the anatomic dissection method.

The fetal period that starts from the 9th gestational week and lasts until birth is the cycle during which the body rapidly grows and the organs mature (Moore & Persaud). Like the other fetal structures, the dimension of calcaneal tendon increases. During the fetal period, parametric

measurements as to the morphological development of calcaneal tendon and the structures constituting it were taken from human fetuses (Tables I, II and III). When we looked back at the previous studies, we did not come across any morphometric studies on calcaneal tendon and the structures constituting it in human fetuses. We have only come across a study conducted on the histological development of calcaneal tendon in human fetuses (Shaw et al., 2008). The studies performed mostly covered the researches on calcaneal tendon in adults (Greenhagen et al.; Kim et al.; Talsnes & Sudmann). Doral et al., in the study they performed on adults, state that the length of calcaneal tendon is at an average of 15 cm (between 11-26 centimeters), the initial width is 6.8 cm (ranging from 4.5 to 8.6 centimeters) and that the termination width is 3.4 cm (ranging from 2.0 to 4.8 centimeters). As the result of the study, it is pointed out that the tendon, after the initial point, becomes gradually narrower towards the end point more prominently in the medial region (Doral et al.). Separately, Kallinen & Suominen compared the tendon width in the elder athletic people with that in the elder sedentary males through the ultrasonographic method. As a result of this comparison, they found that the tendons in the elder athletic ones were much wider. In our study, we took the length, the initial and termination widths of calcaneal tendon throughout the fetal period (Table I). In consequence of our study, we determined that the length of calcaneal tendon increased throughout the fetal period, that the initial region was wide but that the termination point was much narrower than the initial region. However, we ascertained that during the fetal period, the increase in the tendon width within the termination region was much faster than the increase in the width of the initial region (Fig. 2). The result of our study is accordant with the result of the study conducted by Doral et al. According to these results, we came to the conclusion that the length of calcaneal tendon increased right from the beginning of the fetal period, ongoing in the same way in adults, as well. Besides, we deduced that the proximal region of calcaneal tendon, from the outset of the fetal period, began to develop more widely than the distal region, and that it maintained its development in the same way in adulthood, as well. In addition to this, in our study, we also examined the ratio between the length of the leg and the length of calcaneal tendon throughout the fetal period (Fig. 2). We concluded that during the fetal period, calcaneal tendon lengthened more rapidly than the leg (Fig. 2), which we interpreted as a significant gain in terms of the function of calcaneal tendon. Kallinen & Suominen compared the tendon thickness in the athletic male elders with that in the elder sedentary males through the ultrasonographic method. In consequence of their comparison, they could not find any difference between the tendon thicknesses in both groups. Separately, Grechening et al., measured the thickness of the distal region of calcaneal

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tendon in the children, aged 2 months-18 years, through the ultrasonographic method. As the result of the study, they determined that during the childhood between 2 months-18 years, there was an increase in the tendon thickness in the distal region. Besides, in another study, the researchers examined the thickness of calcaneal tendon in children aged below 10 and those aged between 10-17, those aged between 18-30 and in the adults aged over 30 (Koivunen-Niemelä & Parkkola). Consequently, they found out that the tendon thickness in females was slightly thinner than that of the males and that it was proportional to the body height. Furthermore, they ascertained that tendon thickness differed among populations. Different from the other studies, in our research, we measured the thicknesses in the initial and termination regions of calcaneal tendon in human fetuses throughout the fetal period. As a result of the study, we determined that during the fetal period, the initial thickness of calcaneal tendon was more than its termination thickness. Yet, we identified that the increase in thickness in the termination region of calcaneal tendon was faster than that in the initial region of it, starting from the 24-26th gestational weeks (Fig. 2). Moreover, we found out that no difference betwen the genders was observed in terms of tendon thickness, and that it was correlated with the gestational age. Whereas the result of our study was not in accord with the one performed on the male adults by Kallinen & Suominen, it was compatible with the result of the study conducted by Grechening et al. Besides, the result of our study does not show compatibility with that of Koivunen-Niemela & Parkkola in terms of the difference between genders.

In conclusion, our interpretation of the results of this research was that the initial region of calcaneal tendon throughout the fetal period was much thicker, however, the thickness in the termination region of calcaneal tendon continued more rapidly, and that in the postpartum period, the thickness of the initial point slowed down and on the other hand, the termination point continued to get thicker more rapidly and that the thickness in the initial and termination regions of the tendon got balanced during adulthood. Finally, we also concluded that the difference in gender may occur due to age after birth, since it is stated in the studies that the structure of calcaneal tendon changes morphologically and histologically with aging (Strocchi et al., 1991). According to these results, we inferred that in new-born babies and children, the termination region of calcaneal tendon would be thinner and narrower, thus, this would pave the way for tendon ruptures in this region. In adults, on the other hand, we made the interpretation that the tendon width in tendon ruptures would be more important because of the fact that the tendon thickness in the initial and termination regions of the tendon were equally balanced.



Fig. 2. The proportions of the parameters of calcaneal tendon to one another and the proportion of the length of calcaneal tendon to the length of the leg.



Fig. 3. The proportions of gastrocnemius muscle caput mediale and laterale to another.



Fig. 4. The proportions of the parameters of gastrocnemius muscle caput mediale/laterale. Soleus muscle and calcaneal tendon to one another (1/c+f: soleus muscle width / gastrocnemius muscle caput mediale width + gastrocnemius muscle caput laterale width).

As the result of our study, we have not come across any morphometric studies regarding gastrocnemius muscle caput mediale and caput laterale, and soleus muscle in human fetuses. We have only come across one ultrasonographic study researching into the termination positions of soleus muscle in calcaneal tendon in adults and its clinical significance for the KADIR, D.; CEREN, U.; BUSRA, S.; ESRA, K. & ALI, M. M. A Study on the structure and morphologic development of calcaneal tendon and triceps surae muscle in human fetuses during the fetal period and the evaluation of clinical importance of calcaneal tendon. *Int. J. Morphol.*, 33(3):920-929, 2015.



Fig. 5. A) The histological section showing the direction of the fibres of calcaneal tendon. B) The histological section showing the direction of the fibers of calcaneal tendon and the paratendinous tissues.



Fig. 6. A) The histological section showing the soleus muscle. B) The histological section showing the gastrocnemius muscle.

matter involved, and analyzing the effect of growth in children on the geometry of gastrocnemius muscle (Pichler *et al.*; Bénard *et al.*). In our study, different from the others, we took morphometric length, width and thickness measurements of gastrocnemius muscle caput mediale/caput laterale and soleus muscle, which join the constitution of calcaneal tendon in human fetuses (Tables I, II and III). Separately, we examined the proportions of morphometric measurements of caput laterale and caput mediale throughout the fetal period (Fig. 3). Consequently, we determined that during the fetal period, caput laterale and caput mediale developed equally; yet, caput mediale was longer, larger and thicker than caput laterale during this period (Tables I, II and III, Fig. 3). Our interpretation of this result was that caput mediale was more effective than caput laterale in the constitution and function of calcaneal tendon. In our study, we also examined the proportion of the length of soleus muscle to the length of gastrocnemius muscle caput mediale, and the proportion of the width of soleus muscle to the sum of the widths of gastrocnemius muscle caput mediale and caput laterale (Fig. 4). In consequence of the study, we ascertained that during the fetal period, soleus muscle lengthened more rapidly- even if a little- compared to gastrocnemius muscle caput mediale and that soleus soleus was much longer all through that period (Tables I, II and III, Fig. 4). Furthermore, we attained the result that the width of soleus muscle developed evenly (at the same rate) with the sum of the widths of gastrocnemius muscle caput mediale and caput laterale throughout the fetal period (Fig. 4). Our interpretation on these results were that soleus muscle was more effective than gastrocnemius muscle in the constitution and function of calcaneal tendon. We also analyzed, in our study, the proportion of gastrocnemius muscle caput mediale to the length of calcaneal tendon and proportion of the length of soleus muscle to the length of calcaneal tendon throughout the fetal period (Fig. 4). As the result of the study, we determined that calcaneal tendon lengthened more rapidly during the fetal period (Fig. 4). Our interpretation was that this outcome was an important gain in terms of the function of calcaneal tendon.

Moreover, in the histological sections we took from the calcaneal tendon of the fetuses of 15-40 weeks, we determined that the course of direction of the fibers of tendon was longitudinal throughout the fetal period (Fig. 5A-B). In the study conducted by Shaw et al., it was stated that the fibres of tendon were longitudinal until the 38th week of gestation in terms of their course, and that starting from that week, they tended to bend gradually. In adults, however, it is expressed that the fibres of tendon follow a spiral course (Apaydin et al.; Grechenig et al.; Standring et al.). While the results of our study show concordance with that of Shaw et al., it does not show the same parallelism with the results obtained from the adults. Thus, our interpretation of this outcome was that the fibers of tendon continued developing in the wake of the delivery, as well, and that they, later on, transformed into a spiral form as seen in adults. We analyzed the histological structures of calcaneal tendon in the microscopic sections taken from calcaneal tendon. As the result of this analysis, we tried to demonstrate the endotenon, epitenon and paratenon structures of tendo calcaneus (Fig. 5B). Moreover, we histologically analyzed the muscle morphology of soleus and gastrocnemius muscles during the fetal period. As a result of the analysis, we determined that the muscle structure was histologically multipennate and fusiform muscles, respectively, as were similar to the adult counterparts (Fig. 6A-B). That result was also interpreted as the fibers structure of the soleus and gastrocnemius muscles have completed their histological development process during the fetal period.

Calcaneal tendon is the thickest and the strongest tendon in the human body (Apaydın et al.; Doral et al.). Although it is resistable to almost 1 ton of tractive force, it is stated in the studies that it is one of the most frequently ruptured tendons in the body (Apaydin et al.). In developed countries, 50% of all the tendon injuries are reported to occur in calcaneal tendon (Apaydın et al.; Morel et al., 2005; Schweitzer & Karasick, 2000). It is also pointed out in the researches that the muscle tension in calcaneal tendon that has a very important function in the biomechanics of the lower extremity is reported to be approximately 250% as much as the weight of the body (Apaydin et al.; Morel et al.; Schweitzer & Karasick). It is also noted in the researches that this load during running increases 6-8 times more (Apaydin et al.). It is necessary that the anatomy and development of calcaneal tendon, which has quite a major role in human life, be very well known, since it is already stated in the studies conducted on achilles tendon, that with aging, changes occur in the morphology and morphometry of calcaneal tendon (Strocchi et al.). Separately, it is of great importance that the relevant pathologies and anomalies regarding calcaneal tendon during the fetal period be known for the diagnosis and treatment of calcaneal tendon pathologies, such as Haglund deformity and retrocalcaneal bursitis. Therefore, we agree upon the fact that anatomic dissection, during the fetal period, is a proper method for reaching more accurate and reliable results in comparison to the radiological methods. That is why, in our study different from others, detailed morphometric data concerned with fetal calcaneal tendon are emphasized on in a broader range. In order to assess the calcaneal tendon through the obstetric ultrasonography during the intrauterine period, the findings in our study should be evaluated as a pioneering study. Additionally, we are of the opinion that the findings in our study will be beneficial in the evaluation of the pathologies and anomalies related to calcaneal tendon in fetal autopsy materials.

Consequently, we think that in determining the anomaly, pathology and variations with respect to the development of calcaneal tendon during the fetal period, the data obtained in our study will contribute to other studies, such as obstetrics, perinatology, radiology, forensic medicine and phytopathology, and the diagnoses and treatments.

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KADIR, D.; CEREN, U.; BUSRA, S.; ESRA, K. & ALI, M. M. Estudio sobre la estructura y desarrollo morfológico del tendón calcáneo y el músculo tríceps sural en fetos humanos durante el período fetal y evaluación de la importancia clínica del tendón calcáneo. *Int. J. Morphol.*, 33(3):920-929, 2015.

RESUMEN: El objetivo de esta investigación consiste en el estudio del desarrollo morfométrico del tendón calcáneo y las estructuras que se desarrollan con él durante el período fetal humano y evaluar su importancia clínica. El estudio comprendió un total de 102 piernas de fetos (51 fetos humanos: 26 masculinos y 25 femeninos) cuyas edades variaron entre 15 a 40 semanas de edad gestacional, sin presencia de patología externa o anomalía. Los fetos fueron divididos en grupos de acuerdo con las semanas de gestación, trimestres y meses. Luego de realizar mediciones generales externas en los fetos, se procedió a la disección de las piernas. Se midieron parámetros morfométricos correspondientes a los músculos gastrocnemio, sóleo y tendón calcáneo. Los promedios y las desviaciones estándar de los parámetros medidos se determinaron de acuerdo a la edad gestacional de los fetos, en semanas trimestres y meses. Hubo una correlación significativa entre los parámetros medidos y la edad gestacional (p<0,001). No hubo diferencias entre los sexos en términos de parámetros (p>0,005). Todos los resultados obtenidos se discutieron haciendo una comparación entre ellos y los estudios previos realizados. Los datos obtenidos en nuestro estudio serán de utilidad para los médicos en la evaluación del desarrollo del tendón calcáneo y las estructuras que lo constituyen, durante el período fetal, para la utilización de este conocimiento en estudios clínicos y diversas aplicaciones.

PALABRAS CLAVE: Desarrollo fetal; Morfometría; Tendón calcáneo; Anatomía.

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